US Army Medical Research and Materiel

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Methods for Production of Antigens Under Control of Temperature-regulated Promoters in Enteric Bacteria

Features and advantages:

- **Produces inexpensive, easily purified proteins in a broth of tryptone and yeast extract**
- **Increases survival time for cells producing the desired proteins**
- **Chances for contamination are reduced during production process**
- **Does not require large**

This invention provides an improved means for growing *Escherichia coli* bacteria to produce colonization factor antigens (CFAs). Both the naturally occurring and recombinant *E. coli* bacteria encode proteins that are under the control of temperature-regulating promoters. Protein production is improved by growing organisms under the control of the promoters at temperatures less than 35°C until at least the late logarithmic phase. Bacteria are separated from the media followed by purification of the desired proteins.

CFAs confer the ability of pathogenic organisms to colonize in the host organism. Methods previously used for large-scale production of CFAs have been unsatisfactory. Methods using solid media resulted in low yields of the bacteria and CFAs, as well as increased risk of contamination of the desired product. This method uses a liquid broth from which the proteins are easily purified. If the desired protein is not released into the supernatant, it could be obtained from the bacteria by disrupting the membranes.

Antigens produced by the proposed methods could potentially be used as vaccines, as a means for measuring the efficacy of vaccines, as probes to detect antigens from samples, and for biochemical characterization of

Patent Status

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